

In the claims

The following amendments are made with respect to the claims in the international application PCT/EP03/06391.

This listing of claims will replace all prior versions and listings of claims in this application.

1 (Currently amended). ~~Method~~ A method for the identification and quantification of one or more proteins in a sample containing a mixture of proteins, wherein said method comprises the steps of:

- a) [P]providing a sample which contains a mixture of proteins;
- b) [P]providing a reagent for the analysis of peptides ~~which~~ wherein the reagent has the general formula

A-Y-PRG

in which

A constitutes at least one functional group for the reversible, covalent or non-covalent binding to a support material,

Y is a group comprising at least one chelate function for metals being low in isotopes, and

PRG is a reactive group for the selective binding to peptides or other biomolecules to be analyzed;

- c) [C]cleaving the proteins in the sample in order to produce peptides;
- d) [C]coupling the peptides to the reagent of step b);

- e) [S]selecting the peptides labeled in step d) under the employment of a functional group for the reversible, covalent or non-covalent binding to a support material and removal of the unbound peptides;
- f) [R]releasing the bound peptides from the support material and elution from the matrix; and
- g) [D]detecting and identifying the labeled peptides by means of mass spectrometry.

2 (currently amended). ~~Method~~ The method, according to claim 1, wherein the cleavage of the peptides is performed enzymatically or chemically.

3 (Currently amended). ~~Method according to claim 1 or 2~~ The method, according to claim 1, wherein the labeled peptides, after their release from the support material and before their analysis by mass spectrometry, are separated from each other, ~~in particular~~ by means of HPLC.

4 (Currently amended).~~Method according to one of the claims 1 to 3~~ The method, according to claim 1, characterized in that several protein- and/or peptide-containing samples are analyzed together.

5 (Currently amended). ~~Method according to one of the claims 1 to 4~~ The method, according to claim 1, ~~moreover~~ further comprising the sequencing of the labeled peptides.

6 (Currently amended). ~~Method~~ A method for the detection of the relative expression of proteins in a protein-containing sample, wherein said method comprises the steps of:

- a) [P]providing a biological sample which contains proteins;
- b) [P]providing a reagent for the analysis of peptides ~~which~~ wherein the reagent has the general formula



in which

A constitutes at least one functional group for the reversible, covalent or non-covalent binding to a support material,

Y is a group comprising at least one chelate function for metals being low in isotopes, and

PRG is a reactive group for the selective binding to peptides or other biomolecules to be analyzed;

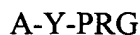
- c) [C]cleaving the proteins in the sample in order to produce peptides;
- d) [C]coupling the peptides to the reagent of step b);
- e) [S]selecting the peptides labeled in step d) under the employment of a functional group for the reversible, covalent or non-covalent binding to a support material and removal of the unbound peptides;
- f) [R]releasing the bound peptides from the support material and elution from the matrix;~~and~~
- g) [D]detecting and identifying the labeled peptides by means of mass spectrometry; and

- h) [M]measuring the relative occurrence of the differently labeled peptides as distinct peaks of ions in order to determine the relative expression of the protein, from which the affinity-labeled peptide is derived.

7 (Currently amended). ~~Method according to one of the claims 1 to 6~~ The method, according to claim 6, characterized in that the arrangement of the groups A, ~~XY~~ and PRG is interchanged.

8 (Currently amended). ~~Method according to one of the claims 1 to 7~~ The method, according to claim 6, characterized in that the labeled peptides are detected by means of a tandem technique[s, like e.g.] selected from the group consisting of matrix-assisted laser desorption/ionization (MALDI), time-of-flight (TOF)-TOF-MS and electrospray ionization (ESI)-MS.

9 (Currently amended). ~~Reagent~~ A reagent for the mass spectroscopic analysis of peptides which has the general formula



in which

A constitutes at least one functional group for the reversible, covalent or non-covalent binding to a support material,

Y is a group comprising at least one chelate function for metals being low in isotopes, and

PRG is a reactive group for the selective binding of peptides or other biomolecules to be analyzed, ~~that shall be analyzed.~~

- 10 (Currently amended). ~~Reagent~~ The reagent, according to claim 9, wherein the arrangement of the groups A, Y and PRG is interchanged.
- 11 (Currently amended). ~~Reagent according to claim 10 or 11~~ The reagent, according to claim 9, wherein the PRG is selected from the group consisting of [a] sulphydryl-reactive groups, ~~an~~ amine-reactive groups and ~~an~~ enzyme substrates.
- 12 (Currently amended). ~~Reagent~~ The reagent, according to claim 11, wherein the PRG is selected from the group consisting of ~~an~~ amine-reactive pentafluorophenyl ester groups, ~~an~~ amine-reactive N-hydroxysuccinimide ester groups, sulfonylhalides, isocyanates, isothiocyanates, active esters, tetrafluorophenyl esters, ~~an~~ acid halides and ~~an~~ acid anhydrides, [a] homoserine lactone-reactive primary amine groups and [a] carboxylic acid-reactive amines, alcohols or 2,3,5,6-tetrafluorophenyltrifluoro-acetates, [a] iodine acetylamide groups, ~~an~~ epoxides, ~~an~~ α-haloacyl groups, [a] nitriles, [a] sulfonated alkyls, ~~an~~ arylthiols and [a] maleimides.
- 13 (Currently amended). ~~Reagent according to one of the claims 9 to 12~~ The reagent, according to claim 9, wherein A is selected from the group consisting of biotin or modified biotin, [a] 1,2-diols, glutathiones, maltoses, [a] nitrilotriacetic acid groups, ~~an~~ oligohistidines and [a] haptens or other reactive reagents allowing for a reversible binding to a support material.

- 14 (Currently amended). ~~Reagent according to one of the claims 9 to 13~~ The reagent, according to claim 9, moreover further comprising a linker between the groups A, Y and/or PRG, which is cleavable in a chemical and/or enzymatic way and/or by exposure to radiation or light.
- 15 (Currently amended). ~~Reagent~~ The reagent, according to claim 14, wherein the linker contains a disulfide group.
- 16 (Currently amended). ~~Reagent according to one of the claims 9 to 15~~ The reagent, according to claim 9, wherein Y is selected from the group consisting of [a] macrocyclic lanthanoid chelate complexes, [a] functionalized tetraaza-macrocycles, [a] polyaza-polyacetic acids, DOTA, [a] DOTA-derivatives, NOTA, [a] NOTA-derivatives, 1,4,7,10,13,16,19,22-octaazacyclotetracosane-1,4,7,10,13,16,19,22-octaacetic acid (OTEC), 1,4,7,10,14-17,20,23-octaazacyclohexacosane-1,4,7,10,14,17,20,23-octaacetic acid (OHEC), EDTA, DTPA-BP, DTPA, DO3A, HP-DO3A and DTPA-BMA.
- 17 (Currently amended). ~~Reagent according to one of the claims 9 to 16~~ The reagent, according to claim 9, wherein the metal bound by the chelate complex is selected from the group consisting of Ag, Al, As, Au, Be, Cd, Ce, Co, Cr, Cu, Dy, Er, Eu, Fe, Gd, Hg, Ho, In, La, Li, Lu, Mn, Na, Nd, Ni, Pb, Pr, Rb, Rd, Sb, Sm, Sn, Tb, Tl, Tm, V, W, Y, Yb and Zn.
- 18 (Currently amended). ~~Reagent according to one of the claims 9 to 17~~ The reagent, according to claim 9, wherein the chelate forming group is labeled with several different metals.

19 (Currently amended). ~~Use of a reagent according to one of the claims 9 to 17 for the detection of~~ A method for detecting peptides in a biological sample and/or for determining the relative expression of proteins in a protein-containing sample wherein said method comprises the use of a reagent for the mass spectroscopic analysis of peptides which has the general formula

A-Y-PRG

in which

A constitutes at least one functional group for the reversible, covalent or non-covalent binding to a support material,

Y is a group comprising at least one chelate function for metals being low in isotopes, and

PRG is a reactive group for the selective binding of peptides or other biomolecules to be analyzed.

20 (Currently amended). ~~Use of a reagent according to one of the claims 9 to 17 for~~ A method for the diagnosis of diseases of an animal, in particular of the human, by detecting the relative expression of proteins in a protein-containing sample taken from the animal wherein said method comprises the use of a reagent for the mass spectroscopic analysis of peptides which has the general formula

A-Y-PRG

in which

A constitutes at least one functional group for the reversible, covalent or non-covalent binding to a support material,

Y is a group comprising at least one chelate function for metals being low in isotopes, and PRG is a reactive group for the selective binding of peptides or other biomolecules to be analyzed.

21 (Currently amended). A [D]diagnostic kit, containing a reagent according to one of the claims 9 to 17 a reagent for the mass spectroscopic analysis of peptides which has the general formula

A-Y-PRG

in which

A constitutes at least one functional group for the reversible, covalent or non-covalent binding to a support material,

Y is a group comprising at least one chelate function for metals being low in isotopes, and PRG is a reactive group for the selective binding of peptides or other biomolecules to be analyzed;

together with further wherein said kit further comprises additional substances and/or enzymes suitable for the detection of peptides in a biological sample and/or the determination of the relative expression of proteins in a protein-containing sample, in particular containing an internal standard.